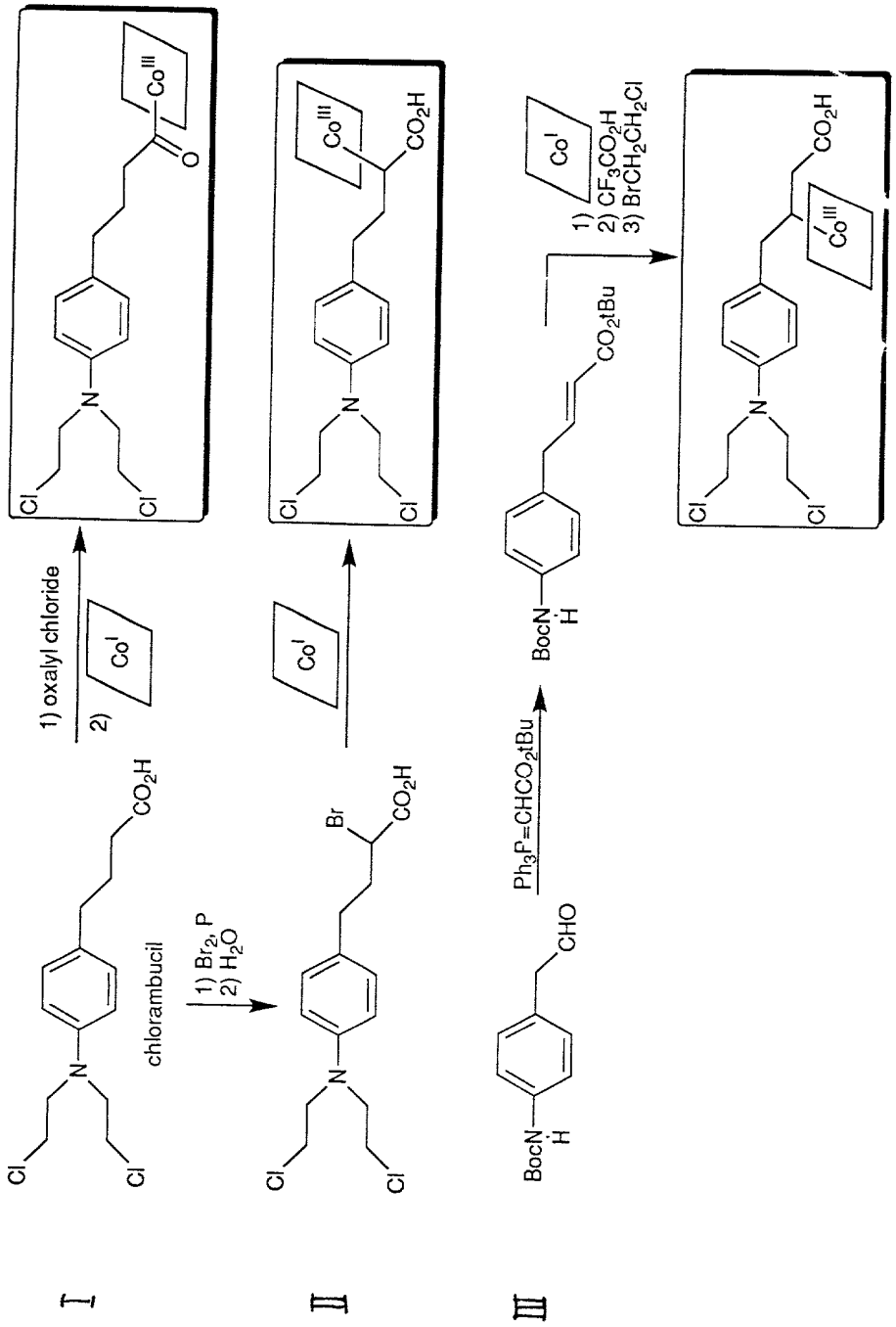
A chlorambucil, ethyl ester-containing bioconjugate can be synthesized by the following methods. When conjugating a drug via a carboxyl group, as in the case of chlorambucil, linking the drug to the cobalamin via a hydroxyethyl tether may be desirable. This can be accomplished by one of two convenient routes, both of which are schematically illustrated below. First, 2-hydroxyethyl-cob(III)alamin can be readily prepared from cob(I)alamin and bromoethanol. Esterification is carried out under standard conditions, i.e. by reaction of a carboxylic acid (chlorambucil) with an alcohol (2-hydroxyethylcob(III)alamin) in the presence of dicyclohexylcarbodiimide (DCC) (or water-soluble derivatives such as EDCI) and a catalytic amount of 4-N,N-dimethylaminopyridine (DMAP) and its hydrochloride salt (DMAP-HCl) in dichloromethane or toluene. Alternatively, the ester-linked conjugate can be prepared by first forming the 2-bromoethyl ester of chlorambucil and then reacting the ester with cob(I)alamin to provide the same product. The reaction schemes (I, II) are shown below. With this mode of attachment, cleavage from the bioconjugate leads to release of the ethyl ester of chlorambucil according to reaction scheme III.


I
II
III
chlorambucil
1) oxalyl chloride
2)
Co'
1) Br2, P
2) H2O
Ph3P=CHCO2tBu
1)
2) CF3CO2H
3) BrCH2CH2Cl
CO2H
CO2tBu
Br
CHO
BocN
H
Cl
N
O
Co'''
Co'

An etoposide-containing bioconjugate can be synthesized by the following method. Etoposide is a semisynthetic derivative of the natural product epipodophyllotoxin that is widely used against a variety of tumors, especially small cell lung carcinoma and germ cell tumors (De Jong et al., 1995). It has also shown considerable promise in the treatment of refractory cases of ovarian and breast cancer. Etoposide appears to function as a topoisomerase II poison.

Etoposide is conjugated to cobalamin, Co[SALEN] and other organocobalt complexes according to the following reaction schemes. Bioconjugates **8a** and **8b** require conversion of the free phenol of etoposide (**3**) to the corresponding chloroformate **17**. Direct acylation with Co(I) gives acylCo(III) derivative **8a**, while treatment with the previously described hydroxyethylCo(III) derivative **18** furnishes carbonate **8b**. This derivative is also available via acylation of **3** with the chloroformate **19** derived from **18**. Preparation of acetal-modified conjugate **8c** may be more challenging. The ethylene acetal of **3** can be hydrolyzed and then the acetal reformed using aldehyde **20a** or dimethyl acetal **20b** (Keller-Jusl et al., 1971) Compound **20a** may also be accessed via careful, selective oxidation of **18**, while **20b** should be available via alkylation of the Co(I) derivative with commerically available bromoacetaldehyde dimethyl acetal. In addition, the acetal of glucose can be formed and then the secondary alcohol of **21** can be glucosylated . Cleavage of **8a** or **8b** either give **3** directly via fragmentative pathways, or furnish products which can undergo eventual hydrolysis to **3**. Trapping with H• following homolysis of **8c** would then furnish **3**.